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Anemia Following Nerve Resection.

By

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There has been much written about the vegetative nervous regulation of the blood. Comprehensive works have appeared too concerning these researches /1-9 /. But we possess less knowledge about the influence of the peripheric nervous system on the blood.

The effect in the leucocyte count resulting from the transection of different peripheral nerves, as well as the changes taking place in the histological pattern of the thus "denervated" bone marrow areas, have been dealt with in a few reports only /10-15 /. The results are often contradictory and based not infrequently on rather doubtful methods. Absolutely nothing is known of the effects of peripheral nervous lesions on the erythrocyte system and on the reticuloendothelial apparatus.

We have systematically studied the influence of an impaired peripheral nerve supply on the blood. The effects of nervous lesions were investigated experimentally on a total of 232 albino rats from the same inbred strain. In repeated tests the feces of the animals proved to be free from helminth ova.

The initial red cell count varied from 7,6 to 9,8 million, the hemoglobin from 12,8 to 17,6 gm per cent and the leucocyte count from 6800 to 18000. Maximum individual spontaneous variation for RBC count was ± 600.000; for Hb ± 8 per cent; for WBC count ± 3000;

As the basic experiment an easily and exactly reproducible procedure was used; the high resection of the sciatic and femoral nerves on

one side. The sciatic nerve was transected between its emergence from the pelvis minor and its division into the tibial and peroneus nerves, whereas the femoral nerve was cut in the line of the inguinal ligament. The operation was performed under ether anaesthesia and aseptic conditions. The wounds healed primarily in every case. By the described procedure the ipsilateral tibia and the distal third of the femur were almost totally deprived of their nerve supply.

Unilateral sciatic and femoral nerve resection has been performed in a total of 147 rats, divided into groups of 6 to 8 animals each. The single groups were hematologically studied for 1 week before operation and for at least 60, often 120 to 180, and in a few cases for 300 days following operation. 5 control rats to each experimental series were maintained under the same environmental conditions and on the same diet as the experimental animals.

One day after the nerve resection a slight leucocytosis /21-24.000/ could be observed with neutrophilia and relative lymphopenia. These changes in the differential white cell count are much the same as those following any kind of surgery. On the 2nd to 3rd postoperative day the leucocyte count dropped to the normal level to rise again on the 6th to 15th postoperative day. At this time the WBC count of 60 per cent of the animals reached 25-40.000 with a definite shift to the left and a marked monocytosis /6-14 per cent/, but the changes in the leucocyte system were never so marked and characteristic as those affecting the erythrocyte system.

The most striking and constant change in the blood cell system was a marked anemia. The development and course of this anemia showed no seasonal or sex variations.

In 67 per cent of the cases the anemia following unilateral sciatic and femoral nerve resection was extreme in degree. The red cell count dropped from the initial 7,6 to 9,8 million to below 3 million. The anemia was hypochromic; The animals tolerated this severe loss of erythrocytes relatively well.

Red cell count and hemoglobin values were in 34 per cent of the rats markedly reduced as early as 3 to 6 days after nerve resection. In 40 per cent of the animals anemia developed on the 6th to 15th, and in 26 per cent on the 15th to 30th post-operative day. Then anemia continued to increase. The time of the maximum fall in the red cell count is shown in Fig. 3.

Fig. 3.

In 37 per cent of the animals, after a few weeks of anemia /usually 30 to 90 days after operation/, the red cell count and the hemoglobin values began to rise and reached or approached the initial levels. This increase was often undulant in type, being interrupted by periods of decline in both values. In 63 per cent of the rats red cell counts and hemoglobin values failed to return to the initial level during the period of observation. Of these 67 animals 38 died of severe anemia. 32 were studied at necropsy. 23 exhibited in internal organs exclusively changes due to anemia, whereas in 5 rats the excessive anemia was associated with paravertebral penumonia and in 4 others with multiple small lung abscesses.

Thus the course of anemia following unilateral sciatic and femoral nerve resection is of 3 types /see Fig. 2./

1./ regenerative type /type R/
2./ undulant type with various outcome /type U_R and U_M /
3./ malignant type, which is mostly fatal. /type M/

Fig. 2.

Table 1 demonstrates the main changes in the red cell count and hemoglobin values of the 167 rats.

Table 1.

An interesting parallelism could be observed between the development, course and severity of the anemia and of the visible trophic changes /edema, erosions, trophic ulcers, mutilations/ of the chronically denervated limb. Thus it seemed [illegible] arising from the area with impaired [illegible] to play the greatest role in the genesis of [illegible] the former

was lege artis amputated at the border of the proximal one third in 8 rats. With this high amputation of the femur the sciatic and femoral nerves were cut at the same level as in the nerve resection experiments, yet none of the 8 amputated rats exhibited appreciable anemia during the 90 days of observation. On the contrary, 10 to 30 days after operation red cell count and hemoglobin value exceeded slightly the pre-operative level in each animal.

The data of the last series prove that the decisive factor in the development of the anemia observed is not the nerve resection proper, but some pathological humoral or neurohumoral effect originating from the almost totally denervated area.

In 6 rats we amputated one hind leg 48 hours after having resected the sciatic and femoral nerves of the same leg. We could not observe any significant lasting change in the blood cell system of these animals either. This experimental series show that within 48 hours after nerve resection no such irreversible changes develop that would give rise to anemia, despite the removal of the denervated area.

In further experiments in 10 rats the resection of the sciatic nerve alone, in 6 rats that of the femoral nerve alone, and in 8 to the resection of the median and ulnar nerves, was performed. Nerve resections were followed by anemia in each experimental group. Anemia and trophic disturbances secondary to sciatic nerve resection were of great severity in nearly the same percentage of experimental animals, as after the combined resection of the sciatic and femoral nerves. This agrees well with the fact that the sciatic nerve has the principal share in the nervous supply of the hind leg. Resection of the femoral nerve, and that of the median and ulnar nerves, respectively, was never followed by trophic disturbances, and, according to this, the fall in red cell count and hemoglobin value was also moderate /Table 2./

Table 2.

It is clear from Table 2, that albino rats develop anemia after

the resection of any major peripheral nerve. There are, however, marked differences in the grade of anemia according to the size of the area supplied by the resected nerve, and thus according to the trophic disturbances resulting from resection. It is also obvious that, unlike in the experimental series with nerve resection, no significant permanent changes in the red cell count developed either in the intact control group, or after the amputation of one hind leg.

The above experiments have shed light on the intrinsic relationship between anemia and trophic disturbances following nerve resections. The profound influence exerted by internal and external environmental factors on the development of trophic disturbances is well known. We tried therefore to find out what role do those factors play in the development and maintenance of the anemia, which are activated by the interference with the nerve supply, and act secondarily on the blood cell system.

It has, accordingly been investigated whether the anemia following nerve resections is due to

1./ changes in the distribution of blood cells,
2./ loss of blood,
3./ infection,
4./ local tissue destruction,
5./ dietary deficiency.

1./ Severity and duration of nerve resection anemia clearly contradict the supposition that the anemia would be simply a sequel to an alteration in the distribution of blood cells. The changes in the differential count and in the bone marrow /dealt with later/ completely exclude this possibility. Nevertheless, even if it is not a decisive factor, the role of any change in the distribution of blood cells had still to be cleared. Therefore repeated blood count studies were made in 6 rats at the onset, peak, and improvement of the [illegible] of the sciatic and femoral nerves is [illegible]

2./ Following the combined resection of the sciatic and femoral nerves, the distal two-thirds of the operated limb are anesthetic. As a result, this area is often object to injury. The excoriations liable to occur are, however, superficial, they bleed little, and bleeding ceases rapidly. The same applies to the mutilations in the parts with trophic disturbances. The blood loss in such cases is so insignificant that it cannot be responsible for the extremely severe and lasting anemia following nerve resections, because

a./ also animals showing not even the slightest excoriation had erythrocyte counts as low as 1 million.

b./ In 6 intact control rats "chronic hemorrhagic anemia" was induced by withdrawing blood 12 times in 34 days. Every time 3 to 5 ml blood was taken by heart puncture. This meant an enormous loss of blood compared to the rats' whole blood volume and, yet, the red cell count did not remain persistently below 3 million unless "bleeding" was repeated every day, or every other day. /Fig.3./

c./ At necropsy no evidence of internal hemorrhage was found in any rat that succumbed to nerve resection anemia.

Fig. 3.

3./ It is common knowledge that areas with impaired nerve supply and trophic disturbances fall easily victim to infection.

a./ The rat, being little susceptible to pygenic bacteria, seldom develops purulent inflammation in the area of trophic ulcers or in adjacent tissues. When such an inflammation was observed animal inoculations were done but there did not occur even the slightest fall in the red cell count of the inoculated animals.

b./ 29 rats were injected 2000 U of crystalline penicillin daily by the subcutaneous route for 6 weeks, and 12 rats 4 mg/100 gm body weight terramycin intramuscularly for 4 weeks after unilateral sciatic and femoral nerve resection. The results of these experiments showed that neither penicillin, nor terramycin treatment could prevent the development of nerve [illegible] anemia.

e./ It has been proved in further experimental series /involving a total of 28 rats/ that Neosalvarsan, in doses adequate to prevent Bartonella anemia /0,015 gm/ 100 gm. body weight, divided in 5 doses /does not inhibit the anemia following sciatic and femoral nerve resection and does not cure the anemia developed. These results exclude the possibility that nerve resection anemia is merely a sequel to a manifestation of a latent Bartonella infection.

f./ No evidence has been found of any latent infection at necropsy of animals with sciatic and femoral nerve resection.

g./ In further experiments aimed at the exclusion of the role of some latent infection, splenectomy and partial hepatectomy were carried out and the inguinal lymph nodes of the nerve-resected leg were removed under aseptic conditions in 9 rats with severe nerve resection anemia / 1 to 2 million RBC/. Aerobic and anaerobic cultures made from these organs yielded negative results. Sterile suspensions were prepared from the same organs with physiological saline and 0,5 ml of the suspension was injected subcutaneously into the inguinal region and further 0,5 ml intraperitoneally to 9 intact control rats. /Fig.4./ The animals developed and fed well during the 1 month of observation, no local changes occurred at the site of injection and no appreciable changes were observed in their blood count. 30 days after inoculation unilateral sciatic and femoral nerve resection was performed in all of the 9 rats. In response to this, each animal developed severe anemia.

Fig. 4.

Thus, all the above experimental series disfavour the role of secondary infection in the genesis of nerve resection anemia.

4./ It has been mentioned formerly that nerve resections often lead to the development of trophic ulcers, deformation and mutilation of the distal part of the limb. In order to elucidate the role of [illegible] factors, extensive tissue destruction [illegible], 0,3 ml of a 10 per cent acid solution under the skin of a hind paw, into the [illegible] and, through a bone resection opening,

into the bone marrow of the tibia. The severe, torpid, purulent ulcers, deep tissue and bone necroses which had developed healed in 6 to 8 weeks. Temporarily a slight fall took place in the red cell count /800.000 to 2,000.000/ and in the hemoglobin level / 7 to 24 per cent /, but in spite of the great tissue destructions none of the rats developed even a moderate lasting anemia.

In other control rats we induced severe tissue destruction by mechanical trauma /two blows from a height of 50 cm with a hammer of 1 kg weight to the lower leg and the paw of one hind limb./ In spite of the extensive opened and closed bone fractures and severe tissue destructions, no appreciable anemia could be observed in any of the animals. On the 1st to 3rd day after trauma a slight fall occurred in both red cell count /700.000 to 1,200.000/ and hemoglobin value /8 to 17 per cent/.

These experimental series prove that local tissue destruction can play any significant role in the genesis or maintenance of the anemia following nerve resection.

5./ The experimental rats were fed both quantitatively and qualitatively the same diet as the controls. After resection of the sciatic and femoral nerves the animals showed no loss of appetite and ate the same food as the controls. This observation has been corroborated in studies on a group of 12 rats, the eaten food of which was carefully weighed for one week before and for 4 weeks after nerve resection. On the basis of these results we could exclude the possibility that some dietary deficiency would be responsible for the development or maintenance of the nerve resection anemia.

The anemia develops rather soon after nerve resections. As the anemia is due neither to hemorrhage, nor to infection, it is probable that it was by increasing the rate of hemolysis that resections induced the relatively rapid fall in red blood cell and hemoglobin level. To elucidate this problem rats with the sciatic and femoral nerves resected on one side were subjected to studies on

1./ changes in reticulocyte count,
2./ the acid and osmotic resistance of erythrocytes,
3./ the behaviour of serum bilirubin,
4./ changes in urobilin excretion.

1./ Nerve resections were usually followed within a few days by marked reticulocytosis /1o to 3o per cent/. The increase in the number of reticulocytes coincided with the fall in red cell count and hemoglobin value. With the aggravation of anemia the reticulocyte count continued to increase, reaching 5o to 88 per cent at the peak of anemia. In the course of improvement, reticulocytosis rapidly diminished and returned to the initial level usually 1 to 3 weeks before the red cell count and hemoglobin level became normal.

When anemia is at its peak pathological young reticulocyte forms appear; these are smaller than the normal reticulocyte and even than the normal red cell, have irregular outlines and surface structure and stain a homogenous deep blue with brilliant cresyl blue. Numerous achromoretic reticulocytes could be found too.

2./ Studies on red cell resistance.

a./ Resistance to acid.

[illegible] nerve resection an increase in the acid resistance of erythrocytes was greatly increased: a considerable proportion of the erythrocytes was not hemolyzed in the acid Türk solution and thus could be counted in addition to the leukocytes. When the red cell count begins to increase, the acid fast forms disappear.

b./ Osmotic resistance.

In operation slight individual variations were only found in the experimental animals, viz.

[illegible] hemolysis at a NaCl concentration of 0.48 to 0.46 per cent
[illegible] " " " " " 0.34 to 0.36 " "

The osmotic resistance of erythrocytes was markedly increased after nerve resections. Parallel with the fall in red cell count and hemoglobin value at first the range of resistance increased. At the peak of anemia minimum hemolysis took place

in 0.32 to 0.42 per cent NaCl, maximum hemolysis in 0.28 to 0.24 per cent and in some cases in destilled water only.

Changes in the acid and osmotic resistance of erythrocytes ran parallel with the changes in reticulocyte count.

Fig. 5

It is seen in Fig. that the curves for the reticulocyte count, and for the acid and osmotic resistence of red cells are the mirror images of the curve plotted from the erythrocyte count. Acid fast red blood cells appear only at the peak of the anemia and reticulocytosis, together with the greatest increase in osmotic resistance and with the widest range of resistance. The conclusion is that the excessive number of reticulocytes is responsible for the increase in osmotic resistance and the acid fast red cells are pathological young reticulocytes appearing at the peak of anemia.

3./ The changes in serum bilirubin level following sciatic and femoral nerve resection were studied in blood samples obtained by heart puncture. The preoperative bilirubin level was 0.2 to 0.4 mg per cent; 3-8 days following nerve resections, indirect serum bilirubin levels sometimes increased by 2 or 3 tenth of mg, but even then did not exceed 0.8 mg per cent. Later the serum bilirubin level decreased in every case parallel with the increase and with the duration of the anemia. Serum bilirubin was usually reduced to unreadable levels, which was also conspicuous by the light grey colour of the serum.

4./ Estimation of urobilin excretion.

The daily urobilin output was estimated for five days before and for 2 or 3 weeks after unilateral sciatic and femoral nerve resection. The estimation was carried out according to Neilmeyer / 16 /. The average pre-operative daily urobilin output of the experimental rats was 0.16 to 0.30 mg/100 g body weight. The so-called "Urobilinausscheidungsindex" /i.e. the mg urobilin falling [illegible] 100 g of circulating hemoglobin/ was found to be 16 to 18, i.e. it varied within similar limits as in humans, though with somewhat higher values. This is obviously due to the fact that erythropoiesis is more active in the rat, than in man.

During the first few days following unilateral sciatic and femoral nerve resection, urobilin output showed a slightly increasing tendency. This transient increase, however, was so slight, that mostly it could not be evaluated at all, considering the great variability of spontaneous daily urobilin output. As anemia increased in severity, urobilin output decreased in all experimental animals, only the "Urobilinmauserungsindex" increased /3o to 6o/ in the presence of anemia.

The typical sequence of changes in urobilin output during nerve resection anemia is illustrated in Fig.

The premortal reduction of the "Urobilinmauserungsindex" seen in Fig. 6 was explained by the fact that the animal had been anuric for 24 hours prior to death.

Fig.

The morphological changes in peripheral blood and bone marrow following nerve resections are summed up briefly in the following:

Nerve resection anemia proved to be hypochromic, occasionally normochromic. Hematocrit values declined after nerve resection parallel with the grade of anemia from the pre-operative 48 to 54 per cent to 9 to 21 per cent and the average Hb saturation diminished from the initial 29 to 32 per cent to 11 to 19 per cent at the peak of anemia. The Price-Jones curve was extremely wide and flat in every case. The average diameter, however, showed hardly any change. Fig. 7 shows the Price-Jones curve of one the experimental rats, before, and 1o days after nerve resection.

Fig.

In view of the extreme aniseocytosis and cellular polymorphism present in nerve resection anemia, average red cell values and thickness values would yield absolutely irrealistic data and for this reason have not been calculated.

Unstained smear made it already obvious how variable the red cells are in shape. The pathological structure of red cells was even more evident in unstained gold-shaded smears, made by the technique of Bessis / 17 /. In Fig. 8 normal and

erythrocytes and in Figs. 9 and 10 these from rats with nerve resection anemia are shown.

Fig. 8
Fig.
Fig.

The normal rat erythrocyte is a biconcave disc of regular, smooth surface /Fig. /. In contrast with this, in nerve resection anemia /Fig. / the erythrocytes are varying in size and shape, exhibit an irregular structure and are unusually flat. It is especially obvious in Fig. that the centre of the red cells is almost empty; these anulocytes are typical in marked hypochromic anemia. Fig. illustrates the remarkably varied and bizarre erythrocyte structures. A few target cells are also present among the most polymorphous anulocytes.

We have studied also the electronmicroscopic appearances of hemolysed red cells of rats with nerve resection anemia, in comparison with erythrocyte membranes of intact control rats and of rats with severe anemia induced by repeated bleeding.

Fig.
Fig.

In contrast with the red cell membranes of the normal and with the reticulocyte stromas which make up about 30 per cent of the hemolysed red cells in chronic bleeding /Fig. /, in nerve resection anemia the red cell membranes exhibit a characteristic appearance: 68 to 84 per cent of them are remarkably rough and dense in structure. A considerable percentage of hemolysed cells is filled with a non-structured mass and the rough granular structure appears only at the periphery /Fig. and /. In smears stained according to Pappenheim the above reticulocytes proved to be identical with basophilic red cells, whereas on treatment with brillant cresyl blue they correspond to the pathological young reticulocytes with irregular structure, which stain a deep homogeneous blue.

Fig.
Fig.

Parallel with the improvement of nerve resection anemia and with the cessation of the extreme reticulocytosis and the normalization of osmotic resistance, the number of stromas with rough structure also diminishes. The number of fine, translucent membranes increases. Some of the membranes contain smaller or bigger "craters", variable in depth and round or irregular in shape. /Fig. /

Fig.

With the disappearence of nerve resection anemia the electron-microscopic picture of the hemolysed red cells and the reticulocyte stromas /present in 1 to 2 per cent/ becomes normal. The changes of the erythrocyte system as observed in peripheral blood at the peak of nerve resection anemia have been listed in Table.

Table.

To study the changes in bone marrow function 3o rats were subjected to serial bone marrow punctures before and after unilateral sciatic and femoral nerve resection.

The pre-operative results agreed well with the data published in the literature. Bone marrow smears made after nerve resection showed profound alterations in bone marrow function. As early as 2 to 6 days following unilateral sciatic and femoral nerve resection the number of reticulum cells was markedly increased. From the 6 th to 15th day erythropoiesis was greatly enhanced, and preponderated over leucopoiesis. Up till 3o to 6o days erythropoiesis often continued to increase and remained high throughout the duration of nerve resection anemia. When this ceased, the cellular pattern of the bone marrow also became normal. In Fig. are illustrated the characteristic changes in the relations of various bone marrow elements.

Fig.

In erythropoiesis there is an maturation. The basophilic normoblasts lymphoid reti-

Erythroblast-phagocytosis is common. Some of the early forms fall victim already in the bone marrow to the reticuloendothelial system.

There is an increase in the number of all the reticulum cell types, especially of the lymphoid reticulum cells. The numerous nuclei without cytoplasm and the numerous Gumprecht shadows indicate, too, the presence of an increase in the number of very young and sensitive lymphoid reticulum cells. Likewise, there is a definite increase in the number of plasmacellular reticulum cells, heparinocytes and macrophages. Lipid-storing macrophages often occur. In some areas blood pigment, whole red cells, nuclear fragments are visible in the reticulum cells.

The myeloid elements show a moderate leftward shift. At the peak of the anemia monocytoid paramyeloblasts, which could not be clearly differentiated, occurred in 4.8 to 6.6 per cent.

Mitoses, often atypical, was frequent in both the red cell and white cell series. The widest variety of chromosome aberrations /appearance of irregular, or too thick chromosomes, fragmentation of chromosomes, rectification of the angle in [illegible] filaments, etc./.

The number of megakaryocytes increased, the "Abschnürung" of the [illegible] was normal.

On account of the limiting values obtained in 30 bone marrow smears, the numerical changes in the cellular bone marrow pattern are presented in Table.

Table.

Discussion.

In the centre of the experiments described is the observation that in albino rats the combined resection of the sciatic and femoral nerves of one limb gives rise to a marked and lasting anemia. We have recognized a parallelism between the chronological course and severity of the anemia, and of the visible trophic changes following nerve resection.

[illegible] relation between nerve resection anemia and the distur-

berce in trophic innervation is well illustrated by our observations after unilateral hind leg amputation. The results of these experiments prove that nerve resection alone does not cause anemia. It is essential for the development of nerve resection anemia that the area with its disturbed nerve supply and trophism remain in the organism.

The parallelism between trophic disturbances and blood cell reactions is shown also by the series, in which resection of different peripheral nerves was performed. According to our findings resection of any major peripheral nerve is followed by anemia, and this anemia is the more severe, the greater the area deprived from its nerve supply.

Considering that trophism means none other than the nervous regulation of tissue metabolism, it is clear why disturbances of tissue metabolism induced by an interference with the nerve supply and the disturbances of a more general nature taking place in the organism /for example anemia/ coincide in time. The whole riddle of individual variations in the development and course of trophic disturbances and blood cell changes finds its explanation in the complex nature of trophic innervation.

In further studies we endeavoured to eliminate the factors caused by the impairment of nerve supply, that may act primarily on the blood cell system and may thus produce anemia.

It has been shown that anemia developing after sciatic and femoral nerve resection was not a sequel to a change in the distribution of blood cells. It is only in the rapid development of anemia that the extreme hyperemia and stasis induced by nerve resection might play some role.

In the next step it has been proved that nerve resection anemia was not due to loss of blood.

Particularly careful studies were conducted to clarify the role of an eventual secondary infection, not only because an area with impaired nerve supply may easily fall victim to infection, but also because nerve resection anemia resembles in some respects the anemia due to infection. In several series of experiments we have eliminated the possibility that local

infection at the site of trophic disturbances, infection by pathogenic agents sensitive to penicillin and terramycin, respectively, or some latent infection /first of all bartonellosis/ would be in the background of the anemia following nerve resection. In spite of the evidence obtained it is not thought improbable that an infection yet unknown, may be involved in the development of nerve resection anemia, although the aforementioned extensive studies apparently contradict this possibility.

On the basis of the parallelism existing between the severity and course of nerve resection anemia and of visible trophic disturbances, it has been suggested that local tissue destruction might play an important role in the genesis and maintenance of the type of anemia under discussion. It has, however, been demonstrated that extensive tissue destruction induced in areas with intact nerve supply, although exceeding in extent and severity the ulcers and mutilations that developed following nerve resection, did not cause appreciable anemia. Thus trophic disturbances following nerve resection are closely related to the anemia, not as if local tissue destruction would lead to the nerve anemia, but because both changes are due to the same fact, lesion of regulation.

As operations on nerves cause not only local disturbances, but also systemic effects, it had to be borne in mind that the change in the general condition of the experimental animal might lead to an eventual dietary deficiency. Quantitative studies of the food ingested revealed that this was not the case.

The relatively rapid fall in red cell count and hemoglobin level following nerve resections suggested the hemolytic origin of the anemia in question. This hypothesis is supported by the extreme reticulocytosis, and by the increase in bone marrow erythropoiesis and by the slight increase of the "urobilinausscheidungsindex". On the other hand, the increased cold test osmotic resistance of the erythrocytes, the low serum bilirubin levels and the fact that no appreciable increase in urobilin output could be proved after nerve resections, contradict the hemolytic origin of the anemia, but do not exclude it. Recent data in anemias of unknown origin / 18 / in cancer

patients /1-21/ and in tumor-bearing animals /22,23/ have indicated a markedly shortened red cell survival time in the absence of marked hemolysis as determined by the usual criteria. It was suggested that red cell survival time is a more sensitive measure of the extent of the hemolytic process. In further experiments we want to study also the changes in red cell survival time in the course of nerve resection anemia.

The experiments have shown that nerve resection is followed by deeprooted changes in the entire blood cell system. In the bone marrow erythropoiesis is definitely enhanced. At the same time, however, maturation is greatly inhibited and signs indicative of pathological changes in both the formation and maturation of erythrocytes are in evidence. The most important manifestation of the disturbance in maturation is the imperfectness, or total absence, of hemoglobin synthesis. As a result of this, at the peak of nerve resection anemia very few orthochromatic erythrocytes are present in the circulating blood. The polychromatic and basophilic red cells, which constitute the overwhelming majority of cells, lose their nuclei at a stage when they contain much ribonucleic acid and hardly any, or no hemoglobin. The pathological structure of these cells has been demonstrated by supravital and gold-shaded light microscopy, as well as by eletron microscopy. In these cells not only the concentration of ribonucleoproteids was increased, but apparently a change took place also in their colloidal state. Our experiments proved that the great increase in the resistance to osmosis and acids observable in nerve resection anemia is due to the extreme reticulocytosis and to the appearance of pathological young reticulocyte forms.

The result of the hematological studies prove that an increase in the activity of the reticuloendothelial apparatus with increased destruction of erythrocytes plays a role in the genesis, and especially in the rapid development, of nerve resection anemia. However, the severity and long duration of the anemia observed are indubitably attributable to a disturbance in erythropoiesis and in maturation.

The hypochromic nature of nerve resection anemia, the extremely low hemoglobin saturation of red cells, the anulocytes, target

cells observed and the remarkably light colour of the serum are signs of a grave disturbance in iron metabolism.

The excessive inhibition of the maturation of erythrocytes, the atypical mitoses, the large numbers of basophilic erythrocytes or of the corresponding reticulocytes with pathological structure indicate that nucleic acid metabolism is gravely impaired.

To sum up in brief how in our opinion nerve resection anemia develops, the following have been concluded from the experiments. After nerve resection grave hyperemia and stasis develop in the almost totally denervated area. As a result, great-masses of erythrocytes are congested in that area and are thus eliminated from systemic circulation. This mechanism may play a role in the rapid fall of red cell count and hemoglobin level following nerve resection.

This impairs circulation and nutrition of the denervated limb. From this area with impaired metabolism humoral or neurohumoral effects influence /through as yet unknown routes, eventually by way of various endocrine glands or internal organs/ changes in reticuloendothelial function, grave disturbances in iron and nucleic acid metabolism and, as a result of all these, persistent anemia associated with extreme reticulocytosis.

Summary.

1./ Unilateral sciatic and femoral nerve resection is followed by a marked and lasting fall in the red cell count and hemoglobin value in the albino rat.

2./ There are different types in the course of anemia.

3./ A definite parallelism has been observed between the course and severity of anemia and of trophic disturbances developing in the limb almost deprived of its nerve supply.

4./ The resection of different peripheral nerves equally results in anemia in the albino rat. The grade of anemia depends on the size of the area supplied by the resected nerve.

5. It is essential in the development of nerve resection anemia that the area with impaired nerve supply, and trophic disturbances remain in the organism. Humoral or neurohumoral effects emitted from this area with impaired metabolism [illegible] factor maintaining the anemia observed.

the distribution of blood cells, nor is it a sequel to loss of blood.

7./ Secondary infections, or manifestations of some latent infection /first of all bartonellosis / are not involved in the development of nerve resection anemia.

8./ Chronic tissue destruction induced in areas with intact innervation causes no anemia.

9./ Anemia following nerve resection is not due to a deficiency in nutrition.

10./ Nerve resections are followed by changes in the entire blood cell system, especially in the formation, maturation and destruction of erythrocytes.

11./ Hematological studies carried out after nerve resections indicate an increased reticuloendothelial activity and the impairment of iron and nucleic acid metabolism.

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